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Determination of active components from selected plants used in traditional medicine in Nigeria by GC-MS screening

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Abstract. *Abu MS, Mashi RL, Lawal JY, Ezekiel EH. 2023. Determination of active components from selected plants used in traditional medicine in Nigeria by GC-MS screening. Cell Biol Dev 7: 23-33.* Herbal products have been utilized extensively in treating various ailments for centuries. Because of the enormous range of functionally relevant secondary metabolites of microbial and plant species, natural products and related structures are critical sources for discovering novel medications. The study investigated the phytoconstituents of extract of selected plants commonly used in traditional medicine in Nigeria, which include: bitter leaf (*Vernonia amygdalina* Delile), ginger (*Zingiber officinale* Roscoe), turmeric (*Curcuma longa* L.), bitter kola (*Garcinia kola* Heckel), Garlic (*Allium sativum* L.) and a mixture of all (turmeric, garlic, ginger, bitter leaf and bitter kola) using GC-MS (gas chromatography-mass spectrometry). GC-MS analysis was performed using Agilent 6890 GC with 59739N MSD and GC-MS equipped with Elite-I fused with silica capillary column (Cpsil 8cb: 30 mm x 25 mm x 0.25 mm). The carrier gas was Helium at a 1.5 mL/min flow rate. The sample was injected at a volume of 1 µL. The results of the analysis confirmed the presence of several compounds. The most prevailing compounds of medicinal value are Hexadecanoic acid, Cis-9,cis-12-Octadecadienoic acid, Squalene, Octanoic acid, Benzene, Pentanoic acid, beta.-Bisabolene, Nonanoic acid, aR-Turmerone, (3R,3aR,7R,8aS)-3,8,8-Trimethyl-6- methyleneoctahydro-1H-3a, Undecanoic acid, 2-Pentadecanone, Benzenedicarboxylic acid, Phytol, Linoleic acid, Dotriacontane, 1,3-Cyclohexadiene, 1,3-Cyclohexadiene, 2,6-Octadienal, 7-Propylidene-bicyclo4.1.0, Butan-2-one, 3-Cyclohexen-1-carboxaldehyde, (E)-1-(6,10-Dimethylundec-5-en-2-yl)-4-methylbenzene, 1-Decanol, Tetratetracontane, Caparratriene, Squalene, 9,19-Cyclolanost-24-en-3-ol. The evaluated plants showed the presence of various biologically active compounds as the scientific background of their use in traditional medicine.

Keywords: *Allium sativum, Curcuma longa, Garcinia kola,* GC-MS analysis, Polyherbal, *Vernonia amygdalina, Zingiber officinale*

INTRODUCTION

Plant products have been used in phytomedicines since ancient times (Cragg and David 2001; Belgica et al. 2021). Any plant part, such as the bark, leaves, flowers, roots, fruits, and seeds, may have active components. Herbal medicines are increasingly popular because they are safe, conveniently accessible, and have lower adverse effects (Ekor 2014). Many plants are less expensive and more accessible to most people, particularly in low-income countries, than modern medication, and there is a reduced rate of side effects following usage (Sofowora et al. 2013). These factors may account for their widespread interest and use. Some studies have documented the therapeutic benefits of some plants. Medicinal plants are the primary source of innovative medications and healthcare items (Ramalingum and Mahomoodally 2014). The extraction and characterization of various active phytocompounds from these green factories have developed certain medicines with high activity profiles (Sasidharan et al. 2011). Indeed, business and public demand for medicinal plants are so high that many therapeutic plants face a significant risk of extinction or loss of genetic diversity (Ekor 2014).

According to Mojab et al. (2003), understanding plant chemical ingredients is desirable since it will be useful in producing complicated chemical compounds. Previous studies have reported the phytochemical compounds of numerous plants. Several studies suggest that secondary plant metabolites may be nutritionally significant and play essential functions in human health (Lavecchia et al. 2013).

Sometimes, due to the synergistic effects, crude extract from medicinal plants is believed to have higher biological activity than isolated compounds (Jana and Shekhawat 2010). Plant phytochemical screening has revealed numerous chemicals, including alkaloids, flavonoids, tannins, steroids, glycosides, and saponins. Secondary plant metabolites are defense mechanisms against predation by many microorganisms, insects, and herbivores (Auwal et al. 2014).

Vernonia amygdalina Delile, belonging to the Asteraceae family, is a popular native vegetable in Nigeria, Uganda, and other African nations. It may be found in various biological zones across Africa and the Arabian Peninsula (Im et al. 2016). Phytochemicals extracted and isolated from *V. amygdalina* include saponins and alkaloids, terpenes, steroids, coumarins, flavonoids, phenolic acids, lignans, xanthones, anthraquinones, edotides, and sesquiterpenes (Farombi and Owoeye 2011).

Ginger (*Zingiber officinale* Roscoe) belongs to the family Zingiberaceae, originating from South East Asia. It

is used in many countries as a spice and condiment to add flavor to food. Besides this, the rhizome of ginger has also been used in traditional herbal medicine (Mashhadi et al. 2013).

Curcuma longa L. is a member of the Zingiberaceae family and is widely cultivated in Asia's tropical regions. Its common name is turmeric root and yellow root. It has been used for centuries as a tonic and treats various ailments such as dyslipidemia, gastrointestinal issues, arthritis, and hepatic diseases (Mazzanti and Giacomo 2016).

Bitter kola (*Garcinia kola* Heckel) is a member of the Clusiaceae family, and Clusiaceae includes several significant fruit and medicinal tree species. Most Clusiaceae grow naturally or semi-domesticated but have been rediscovered as neglected or underutilized crops (Maňourová et al. 2019). Bitter kola is used in African ethnomedicine and traditional rituals.

Allium sativum L. is a member of the Liliaceae family that originated in Asia but is now grown in China, North Africa (Egypt), Europe, and Mexico. Various parts of the plant have long been employed in Iranian and other cultures' traditional folk remedies and as a spice and a food additive (Mikaili et al. 2013).

Plants have been employed as a source of medicine for many diseases since antiquity, owing to their variety and perhaps a rich complement of phytochemical and secondary metabolites (Kennedy and Wightman 2011). A wide range of herbs and herbal extracts contain various phytochemicals with biological activity that can be of valuable therapeutic index. Several phytochemicals have been discovered to have various medicinal properties, which may aid in preventing chronic diseases and metabolic diseases (Tungmunnithum et al. 2018).

In this research, some selected plants (*V. amygdalina, Z. officinale, C. longa, G. kola,* and *A. sativum*) that have been previously and currently being used for herbal decoction in Nigeria were evaluated for phytochemical constituents using Gas-Chromatography Mass Spectrometry (GC-MS).

MATERIALS AND METHODS

Collection and preparation of plant materials

Bitter leaf (*V. amygdalina*), ginger (*Z. officinale*), turmeric (*C. longa*), bitter kola (*G. kola*), garlic (*A. sativum*) were purchased from Wukari L.G.A, Taraba State, Nigeria. It was authenticated in the Department of Biochemistry, Federal University Wukari, Nigeria. The plant materials were thoroughly washed, chopped into pieces, and air-dried for five days before pulverization.

Extraction of plant materials

Exactly 200 g of the pulverized plant material was soaked in 500 mL ethanol for 24 hours at room temperature. Filtrate was filtered through Whatman No. 1 filter paper. After filtration, the extract was concentrated to dryness and kept in a refrigerator.

GC-MS (Gas Chromatography-Mass Spectrometry) analysis

GC-MS analysis was carried out on a Perkin Elmer Turbo Mass Spectrophotometer, which, including the column used, was a Perkin Elmer Elite-5 capillary column (30 m × 0.25 mm) with a film thickness of 0.25 mm composed of 95% Dimethylpolysiloxane. The carrier gas was Helium at a 1.5 mL/min flow rate. One µL sample was injected into the GC. The inlet temperature was maintained at 325°C. The oven temperature was programmed initially at 110°C for 4 min, then increased to 240°C. And after that, it was set to rise to 280°C at a rate of 20°C for 5 minutes. The total run time was 90 min.

The Mass Spectrometry transfer line was maintained at a temperature of 200°C. The source temperature was maintained at 180°C. Gas Chromatography-Mass Spectrometry was analyzed using electron impact ionization at 70 eV, and data were evaluated using Total Ion Count (TIC) for compound identification and quantification. The spectrums of the components were compared with the database of the known component spectrum in the GC-MS library. Measurement of peak areas and data processing were carried out by Turbo-Mass-OCPTVS-Demo SPL software.

RESULTS AND DISCUSSION

The GC-MS chromatograms and the identified compounds in ethanol extract of some selected plants, i.e., bitter leaf (*V. amygdalina*), *g*inger (*Z. officinale*), turmeric (*C. longa*), bitter kola (*G. kola*), garlic (*A. sativum*) and their mixture are presented below:

In Table 1 and Figure 1, 1,3-Cyclohexadiene found in bitter cola has a peak area of 10.04%, while octanoic acid has the lowest peak area of 1.05%. The compound with the longest retention time (30.118 minutes) is Methoxyacetic acid; the shortest is Octanoic acid (5.93 minutes).

The compound with the highest peak area in the extract of bitter leaf is E-11-Hexadecenoic acid with a peak area of 14.00, while the lowest peak area of 0.38% corresponds to Undecanoic acid and Hexadecane, as shown in Table 2 and Figure 2. The retention times for Octanoic acid and Bis(2-ethylhexyl) phthalate compounds were 5.941 and 33.691 minutes, respectively.

Table 3 and Figure 3 show that the compound with the highest peak area in garlic extract is (E)-9-Octadecanoic acid, with a peak area of 16.74%, while Alpha-terpineol has the lowest peak area of 0.28%. (+)-Borneol and Dotriacontane had the shortest and the longest retention times at 5.35 and 29.42 minutes, respectively.

The compound with the highest peak area in the ginger extract was identified as Butan-2- one with a peak area of 12.29%, while the lowest peak area was (E)-1-(6,10-dimethyllundec-5-en-2-y-1)-4- methylbenzene (Table 4 and Figure 4). The longest retention time is 23.594 minutes ((E)-1-(6,10-Dimethylundec-5-en-2-y l)-4-methylbenzene), while the shortest is 5.363 minutes ((+)-Borneol).

Table 1. Identified compounds of ethanol extract of *Garcinia kola* by GC-MS analysis

RT (min)	Name	Mol. Formula	MW (g/mol)	Peak Area%
5.939	Octanoic acid	$C_8H_{16}O2$	144.21	1.05
5.939	Benzhydrazide	$C_7H_8N_2O$	136.15	2.72
12.637	Benzene	C_6H_6	78.11	3.06
12.944	1,3-Cyclohexadiene	C_6H_8	80.13	10.04
13.263	N-Ethyl-p-toluidine	$C_9H_{13}N$	135.2062	2.99
13.616	Cyclohexene	C_6H_{12}	82.14	3.78
15.365	Decanoic acid	$C_{10}H_{20}O_2$	172.26	1.60
16.978	Ar-tumerone	$C_{15}H_{20}O$	216.387	15.53
17.345	Ar-tumerone	$C_{15}H_{20}O$	**216.387**	1.11
17.785	N-Ethyl-p-toluidine	$C_9H_{13}N$	135.2062	2.70
17.850	Sulfurous acid	H_2SO_3	82.07	2.82
20.594	1-Propanol	C_3H_80	60.09	1.12
20.809	Heptadecane	$C_{17}H_{36}$	240.5	0.83
21.360	Didodecyl phthalate	$C_{32}H_{54}O_4$	502.8	1.23
22.361	Heptadecane	$C_{17}H_{36}$	240.47	7.46
22.529	Heptadecane	$C_{17}H_{36}$	240.47	0.81
23.249	Dibutyl phthalate	$C_{16}H_{22}O$	278.34	5.75
23.884	Hexadecanoic acid	$C_{16}H_{32}O_2$	256.4	2.15
26.494	N-[[2-p-Tolylsulfonyl]ethyl]phthalimide			6.02
26.993	Tritetracontane	$C_{43}H_{88}$	605.15	0.20
27.111	10-Octadecenoic acid	$C_{18}H_{34}O_2$	296.46	1.27
27.617	Octadecanoic acid	$C_{18}H_{32}O_2$	280.44	2.00
28.051	Methyl dithio phosphonic acid	$C_2H_7O_2PS_2$	158.17	2.73
28.792	1,8(2H,5H)-Isoquinoline dione	$C_9H_7NO_2$	161.16	1.12
30.021	3,5-Dimethylphenol	$C_8H_{10}O$	122.16	2.53
30.118	Methoxyacetic acid	$C_3H_6O_3$	90.08	1.50

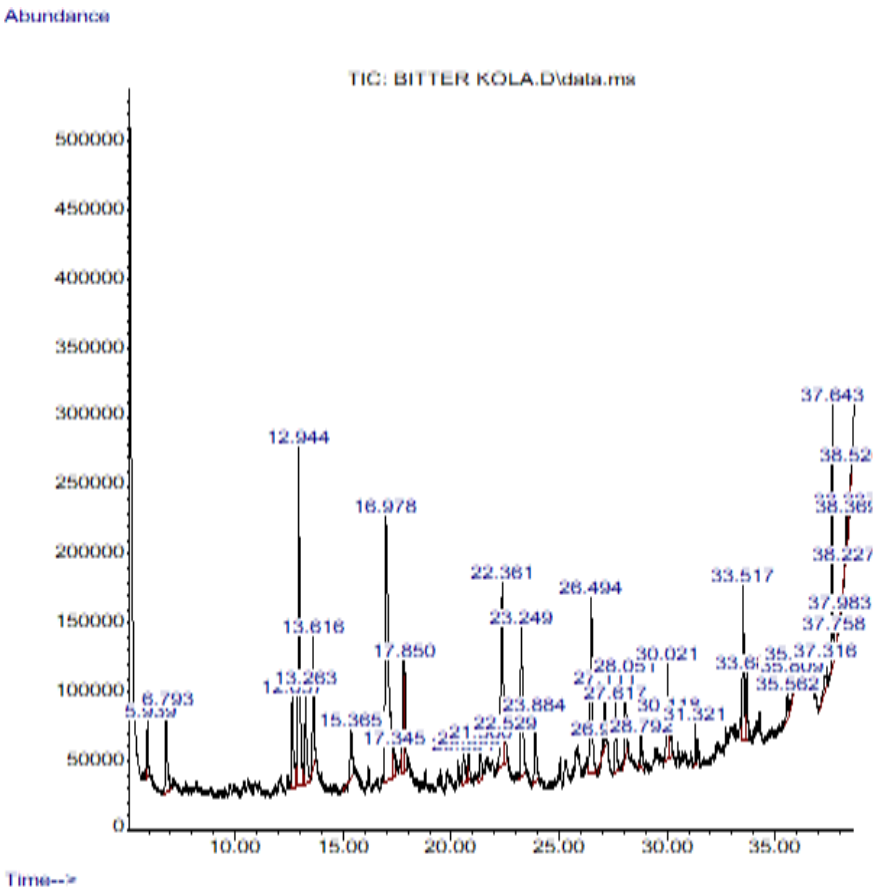

Figure 1. GC-MS Spectrum of ethanol extract of *Garcinia kola* Heckel

Table 2. Identified compounds of ethanol extract of *Vernonia amygdalina* by GC-MS

RT (min)	Name	Mol. Formula	MW (g/mol)	Peak Area%
5.941	Octanoic acid	$C_8H_{16}O2$	144.21	3.06
6.120	Decanal	$C_{10}H_{20}O$	156.26	1.75
12.644	3-Pyridinecarbonitrile	$C_6H_4N_2$	120.11	1.45
12.954	1,3-Cyclohexadiene	C_6H_8	80.13	9.56
13.283	beta.-Bisabolene	$C_{15}H_{24}$	204.35	4.23
13.634	Cyclohexene	C_6H_{10}	82.143	4.39
15.479	Undecanoic acid	$C_{11}H_{22}O_2$	186.29	4.98
19.886	Eicosanoic acid	$C_{20}H_{40}O_2$	312.53	0.40
19.923	Undecanoic acid	$C_{11}H_{22}O_2$	186.29	0.38
20.710	18-Nonadecen-1-ol	$C_{19}H_{38}O$	282.5	0.67
23.917	Hexadecanoic acid	$C_{16}H_{32}O_2$	256.4	11.53
26.238	Phytol	$C_{20}H_{40}O$	296.53	1.72
26.405	4,8-Dioxatricyclo[5.1.0.0(3,5)]octane	$C_6H_8O_2$	112.13	0.95
27.051	9-Octadecynoic acid	$C_{18}H_{32}O_2$	280.4	2.27
27.150	E-11-Hexadecenoic acid	$C_{18}H_{34}O_2$	282.5	14.00
27.653	Undecanoic acid	$C_{11}H_{22}O_2$	186.29	4.48
31.429	1,2,5,6-Di-O-isopropylidene-3-O-me 1thanesulfonyl glucofuranose .alpha.-D-Glucopyranoside			0.67
32.740	Hexadecane	$C_{16}H_{34}$	226.41	0.38
33.524	Bis(2-ethylhexyl) phthalate	$C_{24}H_{38}O_4$	390.55	1.86
33.691	Tartronic acid	$C_3H_4O_5$	120.06	0.90

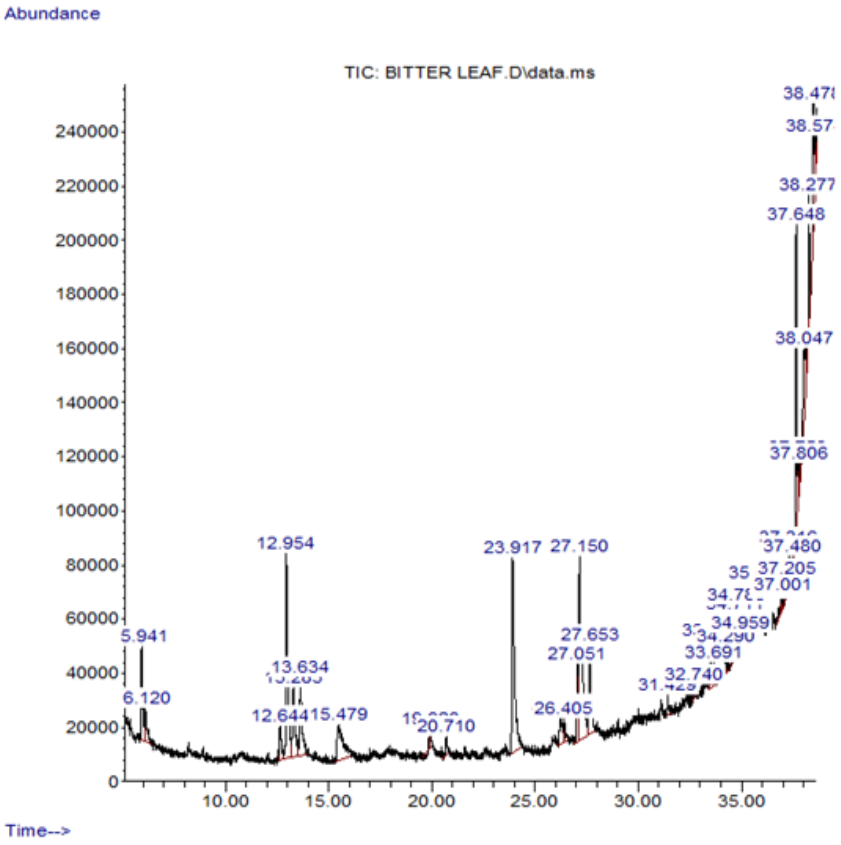

Figure 2. GC-MS Spectrum of ethanol extract of *Vernonia amygdalina* Delile

Table 3. Identified compounds identified of ethanol extract of *Allium sativum* L. by GC-MS

RT(min)	Name	Mol. Formula	M.W.(g/mol)	Peak Area%
5.359	(+)-Borneol Bicyclo[2.2.1]heptan-2-ol	$C_{10}H_{10}O$	154.25	1.14
5.871	.alpha.-Terpineol	$C_{10}H_{18}O$	154.25	0.28
5.940	Octanoic acid	$C_8H_{16}O_2$	144.21	1.31
12.647	Benzene	C_2H_6	78.11	1.66
12.945	Pentanoic acid	$C_5H_{10}O_2$	102.13	1.34
13.274	.beta.-Bisabolene	$C_{15}H_{24}$	204.35	0.97
13.625	(3R,3aR,7R,8aS)-3,8,8-Trimethyl-6-methylene octahydro-1H-3a	$C_{15}H_{24}$	204.35	1.09
15.381	Nonanoic acid	$C_9H_{18}O_2$	158.24	1.27
15.449	Pentanoic acid	$C_5H_{10}O_2$	102.13	0.29
16.988	aR-Turmerone	$C_{15}H_{20}O$	216.31	8.83
19.805	Undecanoic acid	$C_{11}H_{22}O_2$	186.29	1.37
20.839	2-Pentadecanone	$C_{15}H_{30}O$	226.39	0.86
22.536	Pentadecanoic acid	$C_{15}H_{30}O_2$	242.40	1.68
23.258	1,2-Benzenedicarboxylic acid	$C_8H_6O_2$	166.14	1.98
23.880	Hexadecanoic acid	$C_{16}H_{32}O_2$	256.4	10.85
25.770	9,12-Octadecadienoic acid	$C_{18}H_{32}O_2$	280.45	0.80
25.876	7-Hexadecenoic acid	$C_{16}H_{28}O_2$	252.40	2.38
26.141	Phytol	$C_{20}H_{40}O$	296.53	1.05
27.001	Linoleic acid	$C_{18}H_{32}O_2$	280.45	3.67
27.114	(E)-9-Octadecenoic acid	$C_{18}H_{32}O_2$	280.4	16.74
27.625	Octadecanoic acid	$C_{18}H_{32}O_2$	280.44	3.63
29.428	Dotriacontane	$C_{32}H_{66}$	450.87	1.13

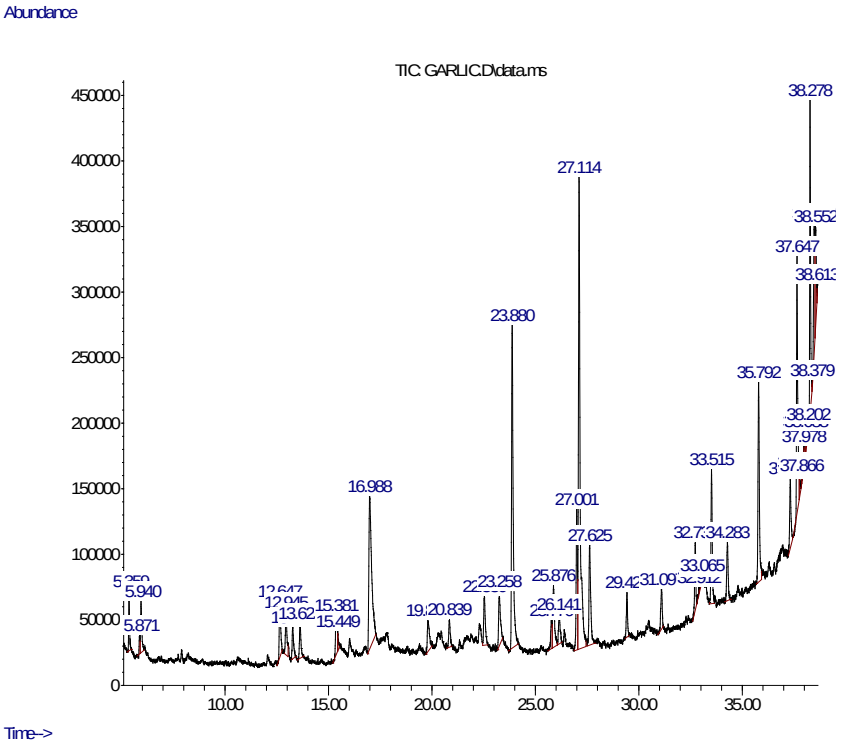

Figure 3. GC-MS Spectrum of ethanol extract of *Allium sativum* L.

Table 4. Identified compounds of ethanol extract of *Zingiber officinale* Roscoe by GC-MS

RT (min)	Name	Mol. Formula	MW (g/mol)	Peak Area%
5.363	(+)-Borneol	$C_{10}H_{18}O$	154.25	0.36
5.942	Octanoic acid	$C_8H_{16}O_2$	144.21	0.38
6.127	Decanal	$C_{10}H_{20}O$	156.26	3.84
12.554	Germacrene D			0.35
12.641	Benzene	C_2H_6	78.11	1.60
12.952	1,3-Cyclohexadiene	C_6H_8	80.12	9.76
13.272	1,3-Cyclohexadiene	C_6H_8	80.12	4.21
13.417	(E,Z)-.alpha.-Farnesene	$C_{15}H_{24}$	204.35	0.43
13.624	Cyclohexene	C_6H_{10}	82.143	4.14
14.265	Cyclohexanemethanol	$C_7H_{14}O$	114.19	0.28
14.351	Cyclohexane	C_6H_{10}	82.143	0.49
14.623	1,6,10-Dodecatrien-3-ol	$C_{15}H_{26}O_2$	222.36	0.41
15.238	cis-p-mentha-1(7)8 dien-2-ol	$C_{10}H_{16}O_2$	152.24	0.79
15.348	Dodecanoic acid	$C_{12}H_{24}O_2$	200.31	0.89
15.777	2,6-Octadienal	$C_8H_{12}O$	152.23	1.12
16.033	7-Propylidene-bicyclo[4.1.0]	$C_{10}H_{16}$	136.23	0.35
16.178	trans-Sesquisabinene hydrate	$C_{15}H_{26}O$	222.37	0.55
16.716	Butan-2-one	C_4H_8O	72.12	12.29
16.953	2-Butanone	C_4H_8O	72.12	6.21
17.486	2-Butanone	C_4H_8O	72.12	1.51
17.636	Butan-2-one	C_4H_8O	72.12	0.32
20.298	3-Cyclohexen-1-carboxaldehyde	$C_7H_{10}O$	110.15	0.81
23.594	(E)-1-(6,10-Dimethylundec-5-en-2-y l)-4-methylbenzene	$C_{20}H_{32}$	272.46	0.32

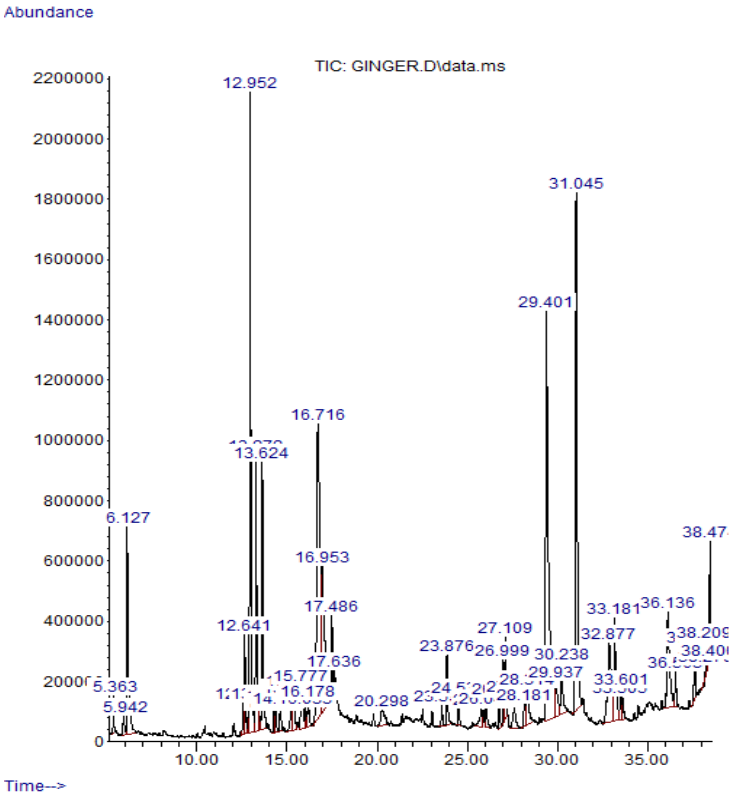

Figure 4. GC-MS Spectrum of ethanol extract of *Zingiber officinale* Roscoe

Table 5. Identified compounds of ethanol extract of *Curcuma longa* L. by GC-MS

RT (min)	Name	Mol. Formula	MW. (g/mol)	Peak Area%
23.945	Hexadecanoic acid	$C_{16}H_{32}O_2$	256.4	1.13
27.177	E-11-Hexadecenoic acid	$C_{18}H_{34}O$	282.46	0.95
32.584	Cyclopropanemethanol	$C_7H_{14}O$	114.19	5.66
32.755	1,6,10,14,18,22-Tetracosahexaen-3-ol	$C_{30}H_{50}O$	426.71	4.41
32.755	Tetratetracontane	$C_{44}H_{90}O$	619.18	4.41
35.823	1-Decanol	$C_{10}H_{22}O$	158.28	0.39
36.370	9,19-Cyclolanost-24-en-3-ol	$C_{30}H_{50}O$	426.7	46.28
37.129	9,19-Cyclolanost-24-en-3-ol	$C_{30}H_{50}O$	426.7	3.48
37.655	Squalene	C30H50	410.73	1.01
38.339	Caparratriene	$C_{15}H_{26}$	207.37	21.64

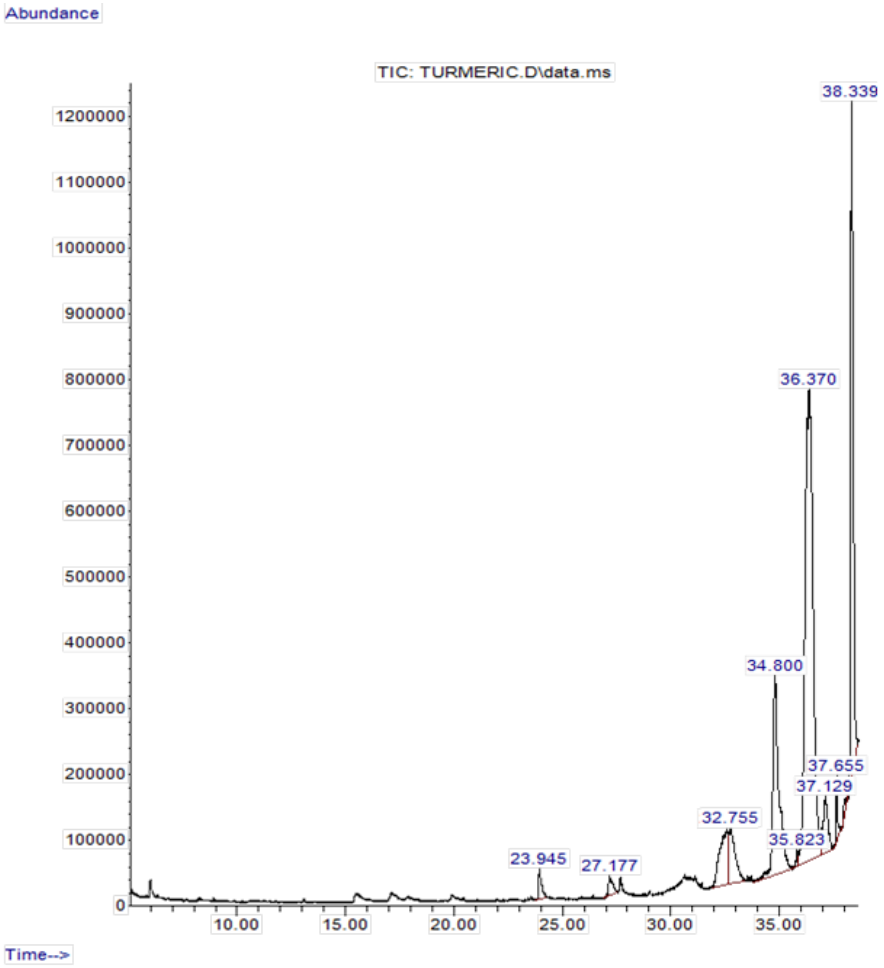

Figure 5. GC-MS Spectrum of ethanol extract of *Curcuma longa* L.

In turmeric, 9,19- Cycloanost-24-en-3-nol has the highest peak area of 46.28%, while 1-Decanol has the lowest peak area of 0.39% (Table 5 and Figure 5). Caparratriene exhibited the longest retention time of 38.339 minutes, while Hexadecanoic acid was eluded at 23.94 minutes.

Table 6 and Figure 6 show that Ar-tumerone has the highest peak area of 12.22% in the mixture of the plants, whereas 1,5- heptadiyne has the smallest peak area of 0.19%. The compound with the highest retention time is 13-Tetradecynoic acid methyl ester (22.542 minutes), while the lowest is 3-dimethyl- (5.39 minutes).

Table 6. Identified compounds ethanol extract of plant mixture by GC-MS

RT (min)	Name	Mol. Formula	MW. (g/mol)	Peak Area%
5.391	3-dimethyl-	C_6H_{12}	84.16	0.27
5.975	Octanoic acid	$C_8H_{16}O_2$	144.21	2.38
5.975	3-methyl-trans-2-Undecen-1-ol	$C_{11}H_{22}O$	70.29	2.38
6.252	methyl- 1-Decanol	$C_{11}H_{24}O$	172.31	0.36
12.649	8-Dimethyl-7-methylene octahydro-1H-3a	$C_{15}H_{22}O_2$	234.33	0.86
12.649	Carbonic acid	H_2CO_3	62.03	0.86
12.954	1,3-Cyclohexadiene	C_6H_8	80.12	5.44
13.286	4,11,11-trimethyl-8-methylene-	$C_{15}H_{24}$	204.35	1.43
13.359	trans-.alpha.-Bergamotene			0.14
13.634	Cyclohexene	C_6H_{10}	82.143	1.83
15.393	10-Bromodecanoic acid	$C_{15}H_{24}$	204.35	3.47
15.534	Octadecanoic acid	$C_{18}H_{32}O_2$	280.44	0.54
15.581	Decanoic acid	$C_{10}H_{20}O_2$	172.26	0.29
17.024	Ar-tumerone	$C_{15}H_{20}O$	216.31	12.22
17.332	Methanone	CO^-	28.010	0.31
17.812	2,4,6-trimethyl-phenol	$C_9H_{12}O$	138.19	3.74
19.401	1,5-Heptadiyne	C_7H_8	92.14	0.19
19.440	2,8-Decadiyne	$C_{10}H_{14}$	134.21	0.44
19.836	Octanoic acid	$C_8H_{16}O_2$	144.21	1.15
22.542	13-Tetradecynoic acid methyl ester	$C_{15}H_{26}O_2$	238.37	0.45

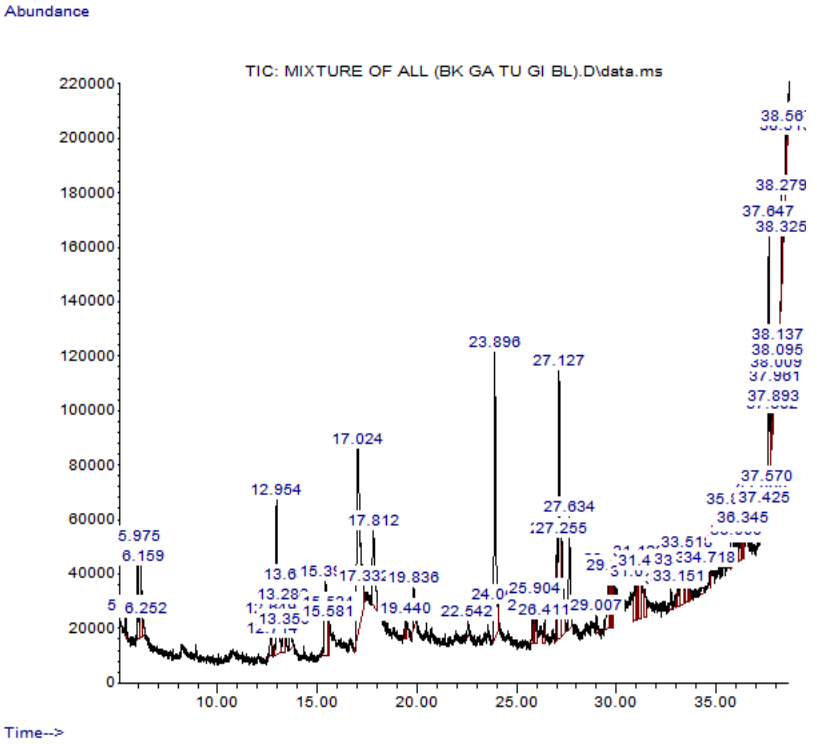

Figure 6. GC-MS Spectrum of ethanol extract of plant mixture

Discussion

Herbs have recently received global attention due to their products' advantages, especially for food and therapeutics (Ekor 2014). Plants bioactive components have various physiological effects, which are principally responsible for their medicinal properties (Altemimi et al. 2017). Essential oils, flavonoids, terpenoids, alkaloids, tannins, phenolic compounds, saponins, and cardiac glycosides are among the bioactive ingredients (Tungmunnithum et al. 2018).

The phytochemicals constituents of some selected plants such as; bitter leaf (*V. amygdalina*), ginger (*Z. officinale*), turmeric (*C. longa*), bitter kola (*G. kola*), garlic (*A. sativum*) and a mixture of all in the ratio of 1:1:1:1:1

(turmeric, garlic, ginger, bitter leaf, and bitter kola) are identified and presented Figures 1-6 and Tables 1-6). The presence of various bioactive compounds in the ethanol extract of the selected plants detected by GC-MS analysis justified the use of the whole plant for various ailments in traditional medicines.

The GC-MS spectrum of *G. kola* ethanol extract revealed 26 compounds (Table 1). The compounds present in the extracts of *Garcinia kola* include: octanoic acid, benzhydrazide, benzene, 1,3-cyclohexadiene, n-ethyl-p-toluidine, decanoic acid, cyclohexene, ar-turmerone, sulfurous acid and didodecyl phthalate, heptadecane, dibutyl phthalate, hexadecanoic acid, n-[[2-p-tolylsulfonyl] ethyl]phthalimide, tritetracontane, 10-octadecenoic acid, Octadecanoic acid, Methyl dithio phosphonic acid**,** 1,8(2H,5H)-Isoquinoline dione**,** 3,5-dimethylphenol, and methoxyacetic acid**.** Ar-turmerone is used in treating neurodegenerative diseases. Benzhydrazide is anticancer and antitumor, while Octanoic acid treats candidiasis and bacterial infections (Zappavigna et al. 2020). Methoxyacetic acid is a hazardous metabolite of ethylene glycol monomethyl ether frequently used as an industrial solvent (Bagchi and Waxman 2008). Janssen et al. (2004) found methoxyacetic acid to increase cellular sensitivity to estrogens, progestins, androgens, and other nuclear receptor hormone receptor ligands. Modulating nuclear receptor transcriptional activity by methoxyacetic acid usually proceeds through activating protein kinases and inhibiting histone deacetylases, which could lead to gonadal toxicities (Blainey et al. 2009).

On the other hand, Camphene (monoterpenes), 1,1,3,3-tetramethyl, Eucalyptol (1,8- cineole), Trans-caryophyllene, and Limonene were absent in the ethanol extract of *G. kola* in this study. Still, they were present in the Edo and Onoharigho (2022) study. On the contrary, results of the GC-MS analysis of the methanolic extract of *G. kola* (Rufa'I et al. 2023) showed the presence of cyclohexane, benzene, and hexanedioic (adipic) acid as present in the ethanol extract of *G. kol*a in this study. However, tridecanoic acid, tetrasiloxane, thymol, pentanone, and silane were absent. The phytochemical variation in the *G. kola* extract could be attributed to the ability of the different solvents to extract bioactive components, which depend on their polarity indexes.

GC-MS analysis of *V. amygdalina* ethanol extract showed 20 identified biologically active compounds (Table 2). The compounds include Octanoic acid, Decanal, 3-Pyridinecarbonitrile, 1,3-Cyclohexadiene, beta.-Bisabolene, Undecanoic acid, Undecanoic acid, Hexadecanoic acid, Phytol, 18-Nonadecen-1-ol. According to Addor (2017), undecanoic acid has antifungal properties, acts as a skin-protective antioxidant, and increases the amount of antioxidants in the skin. E-11-Hexadecenoic acid was the most abundant compound in the ethanolic extract of *V. amygdalina* leaf. This observation was contrary to the report of Alara et al. (2019) on ethanolic extract of *V. amygdalina* leaf, where Phytol was the most prevailing compound while E-11-Hexadecenoic acid was absent.

Table 3 shows the presence of 22 identified compounds as depicted by the GC-MS analysis of the ethanol extract of *A. sativum*. The identified compounds are; (+)-BorneolBicyclo[2.2.1]heptan-2-ol, alpha.-Terpineol, Octanoic acid, Benzene, Pentanoic acid, beta.-Bisabolene, 1,2-Benzenedicarboxylic acid, Hexadecanoic acid, 9,12-Octadecadienoic acid, 7-Hexadecenoic acid, E)-9-Octadecenoic acid, and Octadecanoic acid. In the present study, alpha-terpineol (0.28%) and pentanoic acid (0.29%) were present in modest concentrations in the *A. sativum* ethanol extract. In comparison, (E)-9-octadecenoic acid (16.74%) and hexadecanoic acid (10.85%) were the two predominant components. However, *A. sativum* ethanol extract reported by Park et al. (2017) presented higher concentrations of 2-propenoic acid, trisulfide, allyl trisulfide, 1,3-dihydroxyacetone dimer, acetaldehyde, and dihydroxyacetone. In another report, the extract's main constituents were trisulfide, di-2-propenyl (34.8%), and diallyl disulfide (14.83%) (Gong et al. 2021). The most prevalent non-sulfur components were acetic acid, 2-furan carboxaldehyde, and hexadecanoic acid (Badeli et al. 2022).

The GC-MS analysis (Table 4) of ethanol extract of *Z. officinale* indicates the presence of compounds with diverse biological activity, i.e.; (+)-Borneol, Octanoic acid, Decanal, Germacrene D, 1,3-Cyclohexadiene, (E)-1-(6,10-Dimethylundec-5-en-2-y-l)-4-methylbenzene, (E,Z)-.alpha.-Farnesene, Cyclohexanemethanol, 1,6,10-Dodecatrien-3-ol, and trans-Sesquisabinene hydrate. There is a variation of the compounds in ethanol extract. A previous study by Borekar et al. (2018) showed the presence of 12 major compounds that were identified as Isopropenyl dimethyl, 3-allyl-6-methoxyphenol, thujaketone, 2-butanoic acid, 2-methoxy methyl, hexahydro farnesol, gingerol, tetradecanoic acid, 1-phenyl-3-6-diazohomoadamantan-9-hydrozone, 7-methyl-2-tetradecen-1-ol-acetate, eicosane, androstane and squalene.

The GC-MS analysis of the ethanol extract of *C. longa* determined 10 identified compounds, as represented in Table 5. These compounds are Hexadecanoic acid, E-11-Hexadecenoic acid, Cyclopropanemethanol, 1,6,10,14,18,22-Tetracosahexaen-3-ol, Tetratetracontane, 9,19-Cyclolanost-24-en-3-ol, squalene. Squalene (triterpene) is a phenolic compound found in latex and resins of several plants, with the physiological function that is thought to be a defense mechanism against pathogens that cause human and animal diseases (Scortichini and Rossi 1991; Ezhilan and Neelamegam 2012). In the present study, the GC–MS analyses of the ethanolic *C. longa* extract revealed 46.28% of 9,19-Cyclolanost-24-en-3-ol as the most abundant compound and turmerone (7.14%); however, Abdel-Shafy et al. (2019) reported that the most abundant is turmerone. The presence of squalene reported in the present study was absent in studies by Singh et al. (2011) and Abdel-Shafy et al. (2019).

Lastly, GC-MS analysis of the mixture of the 5 plants showed the presence of some compounds, as shown in Table 6. Some of the compounds are; Methanone, 2,4,6-trimethyl-phenol, 1,5-Heptadiyne, 2,8-Decadiyne, Octanoic acid, 13-Tetradecynoic acid methyl ester, 3-dimethyl-, 3-

methyl-trans-2-Undecen-1-ol, 8-Dimethyl-7 methyleneoctahydro-1H-3. The presence of the compounds in the five plants mixture was known to be used as a flavoring agent, fungicide, pesticide, perfumery, anti-inflammatory, hypocholesterolemic, and cancer-preventive effects (Padma et al. 2019). However, the mixture of the five plants in the ratio of 1:1:1:1:1 does not necessarily increase the horizon of the bioactive components of it compared to the phytoconstituents of the individual plants.

The five plants and their mixture showed various secondary metabolites with relevant biological effects such as antioxidant, anticancer, antifungal, and anti-neurodegenerative activities. However, all the plants and the mixture except *C. longa* contain Octanoic acid, which has been known to have antioxidant activity. *A. sativum* and *Z. officinale* contained (+)-borneol, facilitating digestion and easing pain. Similarly, E-11-Hexadecenoic acid, an antioxidant, was found in *V. amygdalina* and *C. longa*.

Among the compounds present in the *A. sativum*, alpha-terpineol, octanoic acid, (+)-borneol, bicyclo[2.2.1]heptan-2-ol, pentanoic acid, aR-Turmerone, and undecanoic acid exhibit biological activities. Similarly, *Z. officinale* contained octanoic acid, (+)-borneol, germacrene D, and dodecanoic acid, which have biological activity. *Garcinia kola* contained bioactive compounds such as Octanoic acid, benzhydrazide, and Ar-tumerone. *V. amygdalina* contained octanoic acid, undecanoic acid, and E-11-hexadecenoic acid as secondary bioactive metabolites. Meanwhile, E-11-hexadecenoic acid and squalene were found in *C. longa*, Octanoic acid, and 2,8-Decadiyne in the mixture of the five plants as bioactive metabolites. The results revealed that more bioactive compounds were present in *A. sativum* compared to *Z. officinale, G. kola,* and *V. amygdalina*, which had fewer bioactive phytochemicals.

The findings showed the existence of components known to have pharmacological and physiological properties. This research demonstrated that these plants are excellent sources of bioactive chemicals with significant medical value. In conclusion, the phytochemical constituents of ethanol extract of some selected plants, i.e., bitter leaf (*V. amygdalina*), ginger (*Z. officinale*), turmeric (*C. longa*), bitter kola (*G. kola*), garlic (*A. sativum*) revealed the presence of various compounds that may have various biological activities.

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